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EXAMINER	
WOOLWINE, SAMUEL C	
ART UNIT	PAPER NUMBER
1637	

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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/670,701	SU ET AL.	
	Examiner	Art Unit	
	SAMUEL WOOLWINE	1637	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *10 March 2009 and 20 March 2009*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-3,5,7,9-14 and 35-41* is/are pending in the application.

4a) Of the above claim(s) _____ is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-3,5,7,9-14 and 35-41* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☐ The drawing(s) filed on _____ is/are: a)☐ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Status

This application has been transferred to Examiner Samuel Woolwine, whose contact information is listed below.

By way of the supplemental amendment filed 03/20/2009, claims 1-3, 5, 7, 9-14 and 35-41 are pending in the application.

The rejection under 35 U.S.C. 112, 2nd paragraph, made in the Office action mailed 10/10/2008 is withdrawn in view of the amended claims (i.e. the addition of the confounding limitation as dependent claim 35 limited to Raman spectroscopy).

The rejection of claims 1-3, 5, 7 and 9-14 under 35 U.S.C. 103(a) over Woudenberg (U.S. 7,198,900) in view of Hildebrandt (or Kudelski) is withdrawn in view of Applicant's declaration under 37 CFR 1.131 antedating the Woudenberg reference.

The rejection of claims 1-3, 5, 7 and 9-14 under 103(a) over Cleve in view of Dimitrov and Hildebrandt (or Kudelski) is withdrawn in view of the new grounds of rejection over Cleve in view of Dimitrov. Applicant amended claims 1 and 12 in the response filed 03/10/2009 removing the limitations regarding "a signal enhancing surface comprising a salt". As such, Hildebrandt or Kudelski are no longer necessary.

For the same reason, the rejection of claims 1-3, 5, 7 and 9-14 under 103(a) over Singer in view of Urdea, Horn and Hildebrandt (or Kudelski) is withdrawn.

The rejection based on Cleve, Dimitrov and Hildebrandt has been applied to new claims 35 and 36. Therefore, all rejections in this Office action are new rejections

necessitated by Applicant's amendment of 03/10/2009 and supplemental amendment of 03/20/2009.

Applicant's arguments filed 03/10/2009 and 03/20/2009 will be addressed to the extent they apply to the rejections in this Office action.

New Rejections

Claim Rejections - 35 USC § 112

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 1-3, 5, 7, 9-11, 35, 37-39 and 41 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claim 1 recites the limitation "detecting the at least one of the plurality of barcodes bound to the target". There is insufficient antecedent basis for this limitation in the claim. Claim 1 does not recites binding of the at least one of the plurality of barcodes to a target. Claim 1 previously recited such a limitation, but for some reason the amendment submitted 03/20/2009 deleted this limitation, resulting in a claim lacking proper antecedent basis. As the rest of the claims indicated above depend ultimately from claim 1, they are rejected for the same reason. For purposes of examination over the prior art, the examiner will assume claim 1 recites a step of binding to a target (i.e. as recited in claim 41).

Claim Rejections - 35 USC § 103

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

Claims 1-3, 5, 7, 9-14 and 37-41 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cleve et al (Molecular and Cellular Probes 12:243-7, 1998, prior art of record) in view of Dimitrov et al (US 2003/0013091).

The distinction between claims 1 and 12 is that claim 12 further specifies "how" one obtains the plurality of barcodes, and further specifies that a barcode is made up of a nucleic acid template comprising a probe section and a "container" section, wherein the container section comprises two or more different types of tagged oligonucleotides "branched" to the organic molecule backbone of the nucleic acid template. Therefore,

claim 12 is a narrower embodiment of claim 1, and whatever anticipates or renders obvious claim 12 would also anticipate or render obvious claim 1.

With regard to claims 1, 12-14 and 41, Cleve taught a method comprising: (a) obtaining a barcode comprising two or more tags attached to an branched organic molecule backbone (see page 245, columns 1 and 2, where the branched DNA amplifier molecule has 15 branches with four copies of a sequence which bind to labeled probes, where binding of the labeled probes will result in two or more tags attached the branched DNA backbone), (b) binding the barcode to a target (see page 245, column 2, where the probes are hybridized to a target), (C) detecting the barcode bound to the target (see page 246, subheading "Flow Cytometry", where the barcodes are individually detected). Wherein the organic molecule backbone comprises one or more branched nucleic acids (see page 245, column 1 and 2, where branched nucleic acids with 15 branches are used which are organic molecules) and the barcode is detected by a technique of fluorescence spectroscopy (see figure 1, and page 246, column 1, where fluorescence spectroscopy is used to measure the beads). With regard to claim 1, the "branched DNA amplifier molecule" (page 245, sentence spanning columns 1-2) corresponds to the claimed "organic molecule backbone". With regard to claim 12, the "branched DNA amplifier molecule" (page 245, sentence spanning columns 1-2) corresponds to the claimed "nucleic acid template" wherein the "sequence complementary to the preamplifier repeat sequence" corresponds to the claimed "probe section" and the "15 branches" correspond to the claimed "container section".

With regard to claims 2 and 3, Cleve's branched DNA amplifier molecule was single stranded DNA (designed to hybridize with the preamplifier and the probes; see page 245, passage spanning columns 1-2).

With regard to claim 5, Cleve taught the use of a fluorescent dye such as fluorescein (see page 246, column 2, where fluorescein is a fluorescent dye).

With regard to claim 7, Cleve taught branched nucleic acids where the branches were at predetermined locations on the backbone (see page 245, passage spanning columns 1-2).

With regard to claim 9, Cleve taught that the barcode (branched DNA amplifier molecule + probes) binds to a target by way of binding to a preamplifier nucleic acid hybridized to the target. In this manner, the "sequence complementary to the preamplifier repeat sequence" corresponds to the claimed "probe moiety", which clearly mediates binding to the target through the preamplifier nucleic acid.

With regard to claim 11, Cleve taught a nucleic acid target (page 244, "Source of HIV"; page 245, last passage: "8E5/LAV supernate"; the target was HIV-1 nucleic acid).

With regard to claims 37-40, Cleve's barcode is a branched DNA barcode made of labeled oligonucleotides (of known sequence) hybridized to the branched backbone of the "branched DNA amplifier molecule" (page 245, passage spanning columns 1-2).

Cleve does not teach the use of a plurality of barcodes, or that at least one of such barcodes comprised two or more different types of tags on the branched DNA, nor the situation where the number of different barcodes exceeded the number of different types of tags (i.e. labels) (claims 1 and 12). All the labels in Cleve's barcode were

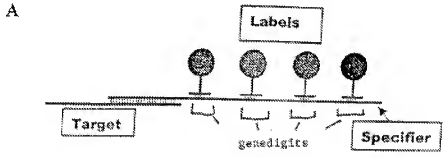
fluorescein. Cleave does not teach generating distinguishable barcodes by attachment of the same tag to different sites along the same backbone (claim 10).

With regard to claims 1 and 12, Dimitrov taught a method of generating "a large number of unique labels of about the same unit signal from just a small number of different labels" (paragraph [0010]). Since a "large number" is clearly larger than a "small number", this clearly implies a plurality of unique labels (barcodes) that is larger than the number of different labels from which the barcodes are made. Dimitrov taught probes comprising "a target specific region and a region containing one or more unique "genedigit" sequences" which "can be specifically bound by a complementary anti-genedigit sequence which can contain a unique label" (paragraph [0011], emphasis provided). This clearly implies a probe with at least two unique genedigit sequences, each able to bind a complementary anti-genedigit with a unique label. This is reemphasized at paragraph [0029] where Dimitrov stated: "The invention also provides unique labels made from combinations of different labels which can increase the number of unique labels substantially." Moreover, Dimitrov taught a specific example at paragraph [0073], where ten unique labels were formed by combining different ratios of two different types of fluorophore. This clearly represents a situation where the number of different types of probes (barcodes) exceeds the number of different types of tags (labels).

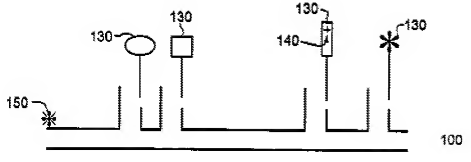
With regard to claim 10, taught a specific example at paragraph [0073], where ten unique labels were formed by combining different ratios of two different types of fluorophore. Based on the text of paragraph [0073], it is clear that these 10 different

labels would comprise the same two different types of fluorophore at different sites along the backbone.

As in the instant disclosure and that of Cleve, Dimitrov's method involved the hybridization of uniquely labeled oligonucleotides to a polynucleotide template. Compare Dimitrov figure 1:



To Applicant's figure 1:



It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to modify the method disclosed by Cleve by designing a plurality of different branched DNA amplifier molecules wherein each different type of amplifier molecule was designed to bind particular ratios of a small number of different tags (labels) in order to derive the benefit taught by Dimitrov at paragraph [0010] of

obtaining a large number of unique labels from just a small number of different labels. In fact, Dimitrov taught at paragraph [0029] that up to 10^{17} different label species could be obtained. Dimitrov taught (again at paragraph [0029]) that "unique labels made from combinations of different labels" would "increase the number of unique labels substantially". Cleve taught multiplexing by using different colored beads (page 244, column 1, last paragraph). Clearly one of skill in the art would have realized that using Dimitrov's idea of unique combinations of a small number of labels to generate a large number of unique labels (by combining different labels in defined ratios) would have represented an alternative to using a different color bead for each unique probe as suggested by Cleve.

Response to Arguments

Applicant's arguments filed 03/10/2009 have been fully considered but they are not persuasive. Applicant argues on page 7 of the response that Cleve does not teach a barcode comprising two or more different types of tags (labels) branched to an organic molecule backbone. This point is not in dispute. The claims are rejected under 35 U.S.C. §103, not § 102. One cannot show nonobviousness by attacking references individually where the rejections are based on combinations of references. See *In re Keller*, 642 F.2d 413, 208 USPQ 871 (CCPA 1981); *In re Merck & Co.,* 800 F.2d 1091, 231 USPQ 375 (Fed. Cir. 1986).

Applicant indicates figure 1 of Tsongalis (Am J Clin Pathol 126:448-53, 2006) as representing the structure of the probes described by Cleve. The examiner agrees this is a fair representation of the structure of Cleve's probes. Applicant argues on page 8 of

the response that Cleve's suggestion to use different color beads for the purpose of detecting different targets in the same assay (see Cleve page 244, column 1, last paragraph) would not have provided a motivation to use different combinations of labels as described in Dimitrov. The examiner respectfully disagrees. Clearly what Cleve was interested in was detecting multiple different targets in a single assay. Dimitrov's idea to make a large number of unique probes by using various combinations of a small number of different labels is totally relevant to this goal. Dimitrov stated at paragraph [0027]: "Enough labels are generated by this method so that each analyte in a complex mixture can be uniquely bound by a label and thus identified."

Applicant states at the top of page 9:

> Each of Cleve's branched DNA produces a unique signal that is distinguishable from other branched DNA of Cleve by tailoring the preamplifier, the number of amplifier molecules and the number of label probes hybridized to the amplifier molecules. In short, with so many degrees of freedom available to design each unique branched DNA of Cleve, persons of ordinary skill in this art would simply have had no motivation to further use different types of labeled probes (tags) in the branched DNA of Cleve.

The examiner does not see anywhere in Cleve's disclosure a discussion of this sort (i.e. tailoring the preamplifier, the number of amplifier molecules and the number of label probes to produce unique signals).

Applicant further argues:

> Applicants respectfully submit that persons of ordinary skill in this art would have interpreted Dimitrov's teaching that "Several unique combinations of labels can be formed using branched nucleic acids (see page 7, paragraph 0057)" as applied to Cleve to mean that the branched DNAs of Cleve could have many different distinguishable signals by tailoring preamplifier, the number of amplifier molecules and the number of label probes hybridized to the amplifier molecules. Persons of ordinary skill in this art would not have interpreted Dimitrov's teaching as a suggestion to modify Cleve's branched DNA to include different types of labeled probes (tags) in the branched DNA of Cleve.

As stated above, Cleve's disclosure does not discuss tailoring the preamplifier, the number of amplifier molecules and the number of label probes to produce unique signals. This is merely argument of counsel, which does not suffice for evidence (MPEP 2145(I)). There is no basis for the conjecture that one of ordinary skill in the art would have adapted Cleve's method in view of Dimitrov's disclosure by "tailoring preamplifier, the number of amplifier molecules and the number of label probes hybridized to the amplifier molecules", as neither Cleve nor Dimitrov discuss such a concept.

Claims 35 and 36 are rejected under 35 U.S.C. 103(a) as being unpatentable over Cleve et al (Molecular and Cellular Probes 12:243-7, 1998, prior art of record) in view of Dimitrov et al (US 2003/0013091) as applied to claims 1-3, 5, 7, 9-14 and 37-41 above, and further in view of Hildebrandt et al (J. Phys. Chem., 1984, Vol.88, pp.5935-5944, prior art of record) and Maniatis et al (Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory, 1982).

The teachings of Cleve and Hildebrandt have been discussed. These references did not teach or suggest detecting the barcodes by Raman spectroscopy, wherein the barcodes are proximately located to a signal enhancing surface comprising a salt from among those recited in the claims.

Hildebrandt demonstrated that the use of a signal enhancing surface in proximity to label during Raman spectroscopy. Hildebrandt demonstrated the use of various anions (Cl-, I-, Br-, F-, and SO_4^{2-} in the form of salts, i.e. HCl, NaCl, or KCl, see abstract and pg.5937, right column, 2nd paragraph from bottom) to enhance the signal of a fluorescent dye (Rhodamine 6G) in proximity to a signal enhancing surface (i.e. colloidal silver) 2-100 fold (see Figures 2 and 3, and Table 1).

According to Hildebrandt, the concentration of Cl^- required for the Raman signal to saturate (i.e. reach 90% of its final value) was 7.5×10^{-4} M (page 5937, column 2, first three full paragraphs; see also table 1). Hildebrandt disclosed that at this Cl^- concentration, the Raman signal was amplified about 170 times over the anion-free state (page 5937, column 2, 2nd full paragraph and see table 1, where "166" is the "about 170"). It is noted that 7.5×10^{-4} M is 0.75 mM.

Page 387 of Maniatis describes a hybridization solution (item 6). This solution calls for 6X SSC. The recipe for 20X SSC is found on page 447: 175.3 g of NaCl per liter. As the formula weight of NaCl is 58.44 g/mol, the concentration of Cl^- in 20X SSC would be 175.3 g ÷ 58.44 g/mol = 3 M. So a solution of 6X SSC, as in the hybridization solution described on page 387, would be 6/20 of 3 M, or 0.9 M, which is 900 mM. That is, the concentration of Cl^- in the hybridization solution of Maniatis is 900 mM, which is

more than enough to produce the maximal Raman signal discussed in Hildebrandt. This provides a reasonable expectation of success that adding a small amount of Cl^- to a hybridization reaction would not have negatively impacted upon the hybridization.

It would have been *prima facie* obvious to one of ordinary skill in the art at the time the invention was made to modify the method suggested by the combined teachings of Cleve and Dimitrov by using Raman spectroscopy during detection where the barcodes are proximately located to a signal enhancing surface in the presence of salt, because Raman spectroscopy was a conventional detection method at the time of the invention for detection of various labels, as demonstrated by Hildebrandt. Furthermore, Hildebrandt demonstrated how the application of a salt to the surface of a signal enhancing surface can increase signals during Raman spectroscopy. It would have been obvious to one skilled in the art to make this modification in order to increase the signal.

Response to Arguments

Applicant's arguments filed 03/20/2009 have been fully considered but they are not persuasive. Applicant argues on page 9 of the response that one would not have been motivated to use the Raman spectroscopy with enhancing salt because Cleve's branched DNA labels already had a large signal to noise ratio. This argument is not persuasive. If there was a means known in the art to improve a signal, it would have been obvious to make use of such means.

Applicant also argues that addition of salt to the hybridization buffer would change the buffer conditions which would change the specificity, which in turn would

result in high levels of false positives or false negatives. Firstly, Applicant has failed to explain how increasing the salt concentration in a hybridization buffer would result in both false positives and false negatives. It was known in the art that increasing salt concentration promotes hybridization. So, given a high enough salt concentration, one could argue that false positives might occur. Secondly, according to Hildebrandt, the concentration of Cl^- required for the Raman signal to saturate (i.e. reach 90% of its final value) was 7.5×10^{-4} M (page 5937, column 2, first three full paragraphs; see also table 1). Hildebrandt disclosed that at this Cl^- concentration, the Raman signal was amplified about 170 times over the anion-free state (page 5937, column 2, 2nd full paragraph and see table 1, where "166" is the "about 170"). It is noted that 7.5×10^{-4} M is 0.75 mM.

Cleve stated of the hybridization buffer (page 245, beginning of last paragraph):

> A hybridization buffer was prepared by adding the following at the indicated final concentrations to Lysis Diluent (Chiron HIV 1.0 kit): 4 mg ml^{-1} proteinase K, 4·5% SDS, 25 mM EDTA, 20 µg ml^{-1} tRNA and 3·8% dextran sulfate (Pharmacia 500k).

While it cannot be determined what the salt concentration in the "Lysis Diluent (Chiron HIV 1.0 kit)" was, if any, it is asserted that 0.75 mM Cl^- is well below the salt concentration in standard hybridization buffers. Evidence for the assertion is found in Maniatis et al (Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory, 1982). Page 387 of Maniatis describes a hybridization solution for Southern blotting (item 6). This solution calls for 6X SSC. The recipe for 20X SSC is found on page 447: 175.3 g of NaCl per liter. As the formula weight of NaCl is 58.44 g/mol, the concentration of Cl^- in 20X SSC would be 175.3 g ÷ 58.44 g/mol = 3 M. So a solution of

6X SSC, as in the hybridization solution described on page 387, would be 6/20 of 3 M, or 0.9 M, which is 900 mM.

In addition, 0.75 mM Cl^- is a far lower Cl^- concentration than found in the 0.1X SSC (standard saline citrate) added to the hybridization prior to pelleting the beads (even accounting for the dilution of the 200 µl SSC by the 50 µl of hybridization reaction; see Cleve page 245, column 2, last paragraph).

Therefore, adding a mere 0.75 mM Cl^- to a hybridization solution as described in Cleve (assuming that Cl^- was not already present in at least this amount) would not have been expected to result in false positive hybridization.

Conclusion

Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to SAMUEL WOOLWINE whose telephone number is (571)272-1144. The examiner can normally be reached on Mon-Fri 9:00am-5:00pm.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on (571) 272-0782. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

/Samuel Woolwine/
Examiner, Art Unit 1637

/Young J Kim/
Primary Examiner, Art Unit 1637